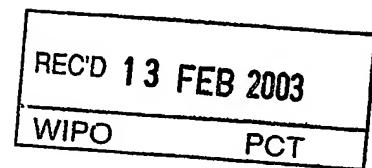




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I, JULIE BILLINGSLEY, TEAM LEADER EXAMINATION SUPPORT AND SALES hereby certify that annexed is a true copy of the Provisional specification in connection with Application No. PS 0145 for a patent by BIOENERGY AUSTRALIA LTD as filed on 25 January 2002.

WITNESS my hand this
Third day of February 2003

JULIE BILLINGSLEY
TEAM LEADER EXAMINATION
SUPPORT AND SALES



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PROVISIONAL SPECIFICATION

Invention Title: "Method of Biomass Development"

The invention is described in the following statement:

"METHOD OF BIOMASS DEVELOPMENT"

FIELD OF THE INVENTION

The present invention relates to a method for generating biomass. In particular, it provides methods of cultivating mixed species tree plantations capable of 5 providing plant material for general biomass demands as well as hardwoods suitable for application in building, furniture etc.

BACKGROUND ART

Due to the rapid increase in the world's population, there is a growing need for timber for building and furniture; woodchips for paper production; and, more 10 recently, as an alternative renewable fuel source to replace fossil fuels. Demand for forest products is increasing, while large areas of forested land are being lost or degraded and timber harvest is being restricted in many of the world's natural forests.

Tree plantations, which are financially very attractive in many locations, offer the 15 potential for meeting large portions of the world industrial wood needs. The supply of plantation-grown wood reduces the pressures and disturbances on old growth forests and natural timber stands. This is possible because the very high productivity of plantation forests requires less area to produce industrial wood. The use of plantations managed for timber production must increase to meet the 20 world's increasing demand for wood and fibre from a reduced land base. Intensive management of plantation forests is perhaps the only way to meet the increasing demand for forest products and still reserve large areas of native forests for conservation and preservation purposes.

Many of the traditionally desirable plantation woods do not, however, grow fast 25 enough to meet demands. Trees such as poplar (with its 'short' rotation period) require 20 years before they are mature enough for harvest. Hoop or exotic pines, which are often used for good-quality sawlogs or ply logs, take 25 to 45 years to mature, while eucalyptus sawlog or pole plantations may require rotations of 40 to 80 years. Even eucalyptus wood for pulping may take 15 years for a 30 rotation.

Despite the pressure to increase wood production from traditional timber plantations, it has been found that high density seedling planting has a negative impact on tree growth and development. Excessive tree density reduces individual tree diameter, which significantly impacts long-term plantation development. This growth loss can be expressed as reduced commercial wood yields and lower value products.

Initial planting densities typically range from 750 to 2500 stems/ha depending on management goals. Close spacing assists in the development of straight stems with fine branching but has a penalty in the development of stem size. Culling or thinning allows the best trees to develop but commonly thinnings may be cut for low-value products or may be wasted.

Plantation trees are typically planted as seedlings, having been grown for a year in a nursery. The cost of seedlings is much higher than that of seeds and planting out is labour intensive or requires specialised machinery.

During establishment of a seedling timber plantation and to a lesser extent over the life of the trees, the trees are extremely vulnerable to nutrient deficiencies as a result of poor soils and out-competition by weeds. Plantations thus require extensive weed control measures and regular application of suitable fertilisers. Problems are also encountered with pests and diseases, due to the vulnerable nature of monocultures. All of these factors can result in reduced growth and timber yield and poor quality timber.

In addition to the aforementioned problems, some highly desirable timber tree species, when grown in a plantation, require protection from the wind in order to grow and develop the tall, straight boles essential for the production of high-quality timber. One way around this problem is to first plant a 'nurse' plantation and then, a few years later, interplant with the desired timber species. This second species will then have the protection of the nurse trees and, just as importantly, the competition which will force it to grow tall and straight. After several years the nurse trees will be removed to allow full growth of the desired high value trees. This, however, is an expensive process and often economically unviable. Some desirable tree species have traits, such as allelopathic

tendencies, which make them difficult to grow in closely spaced monoculture plantations.

Further, many high value timber species cannot be grown in specific regions and sites due to unfavourable environmental conditions such as rainfall, temperature, 5 salinity, soil erosion or a high water table. This limits the range of sites that can be utilised for timber plantations and reduces the potential of timber plantations to reclaim sites such as degraded marginal agricultural land.

Thus, a range of problems have been encountered in meeting the growing demand for plantation timber, which cannot be met by traditional methods of 10 planting and harvesting.

General background

Those skilled in the art will appreciate that the invention described herein is susceptible to variations and modifications other than those specifically described. It is to be understood that the invention includes all such variation and 15 modifications. The invention also includes all of the steps, features, compositions and compounds referred to or indicated in the specification, individually or collectively, and any and all combinations or any two or more of the steps or features.

The present invention is not to be limited in scope by the specific embodiments 20 described herein, which are intended for the purpose of exemplification only. Functionally equivalent products, compositions and methods are clearly within the scope of the invention as described herein.

As used herein the term "derived" and "derived from" shall be taken to indicate that a specific integer may be obtained from a particular source *albeit* not 25 necessarily directly from that source.

Throughout this specification, unless the context requires otherwise, the word "comprise", or variations such as "comprises" or "comprising", will be understood to imply the inclusion of a stated integer or group of integers but not the exclusion of any other integer or group of integers.

Other definitions for selected terms used herein may be found within the disclosure of the invention and apply throughout. Unless otherwise defined, all other scientific and technical terms used herein have the same meaning as commonly understood to one of ordinary skill in the art to which the invention belongs.

5 DISCLOSURE OF THE INVENTION

In the work leading up to the present invention, the applicant sought to develop novel systems for generating large volumes of biomass of high quality and value. Typically, when plants are grown at high density individual plants become suppressed and die because of the competition between the plants. The 10 applicant has identified that plants within the genus *Casuarina* can be grown at high density with little or no detriment to growth or quality of the biomass generated. Such a result is especially significant as it provides a means to generate significant amounts of biomass in a relatively short time period.

Accordingly, in one embodiment the present invention provides a method of 15 cultivating biomass, comprising the steps of:

- (a) Selecting at least a plant variety from the plant genus *Casuarina* capable of growth at a density in excess of about 5,000 stems per hectare; and
- (b) Cultivating the plant variety at a density in excess of about 5,000 stems 20 per hectare.

While this method demands that *Casuarina* plants be grown at a density in excess of about 5,000 plants per hectare, it will be appreciated that the greater the density of the plantings, the higher the volume of biomass that may be recovered from the plantings. In this respect, plantings of greater than 10,000 plants per 25 hectare may be employed in the method of the invention. More preferably, *Casuarina* plants are grown at a density in excess of 20,000 plants per hectare. In a highly preferred form of the invention, *Casuarina* plants are grown at a density between approximately 40,000 and 60,000 plants per hectare.

Where *Casuarina* plants are grown at a density in excess of about 5,000 plants 30 per hectare the plantings should be harvested at relatively regular intervals.

Harvesting times of the plantings will vary depending on the particular variety employed in the method as well as factors such as the rate at which the variety matures and the amount of biomass generated per plant. Preferably, the plantings are harvested every 2 to 4 years, with a 3-year harvesting cycle being 5 highly preferred.

There are several advantages of growing *Casuarina* plants at high density. Those advantages include:

- (i) They grow rapidly, providing shelter to other plant species grown in their proximity;
- 10 (ii) When *Casuarina* plant are grown in proximity to other species, the shelter they provide ensures the other species grow straight and tall;
- (iii) They can be grown directly from seed;
- (iv) They can be repeatedly coppiced and harvested; and
- 15 (v) They have the capacity to form symbiotic root nodules that are capable of fixing atmospheric nitrogen, which facilitates growth of other plants.

Methods for selecting *Casuarina* plant varieties for use in the present invention will be known to those experienced in the horticultural field. As an example only, the selected species may be grown at a desired density in test plots and 20 examined for detrimental effects to plant growth. Preferably, the plant variety is selected from the group comprising: *Casuarina cunninghamiana*, *Casuarina glauca* or *Casuarina obesa* or a hybrid developed from these varieties.

Procedures for developing hybrid plant varieties are well known and are extensively described in the literature. In a preferred form of the invention, the 25 plant variety selected is a hybrid generated by crossing *Casuarina cunninghamiana*, *Casuarina glauca* or *Casuarina obesa* with one of the other aforementioned species. Most preferably, the hybrid variety is generated by crossing *Casuarina cunninghamiana* and *Casuarina glauca*. Hybrids between *Casuarina cunninghamiana* and *Casuarina glauca* have many desirable attributes, 30 some of which include: the hybrid produces very straight, tall stems on a wide range of sites including those where rainfall is moderate and access to groundwater is limited. The hybrid has the potential to be grown in more saline

situations than *Casuarina cunninghamiana* and, from preliminary observations of growth, is able to grow more vigorously on a wider range of sites. It exhibits a high degree of salt tolerance, is suitable as biofuel, exhibits nitrogen fixing capacity, can be direct seeded and coppiced.

5 While a wide range of methods exist for producing hybrid plant varieties, *Casuarina* species are dioecious. Accordingly, to produce *Casuarina* hybrids from seed, special seed orchards are required to generate the seed. Thus, according to an embodiment of the invention there is provided a method for producing *Casuarina* hybrid seeds comprising:

- 10 (a) Growing a first *Casuarina* species to sexual maturity and selecting plants of that species that have a phenotype of female fertility;
- (b) Growing a second *Casuarina* species to sexual maturity and selecting plants of that species that have a phenotype of male fertility;
- (c) Allowing cross-pollination between the female plants with mature pollen from the male plants;
- 15 (e) Raising the female plants to produce hybrid seeds having genetic material from both parents, and
- (f) Harvesting the hybrid seeds.

According to this method the first and second species are preferably grown in the
20 same orchard in alternate rows. When the plants of the first *Casuarina* species near sexual maturity, all the males of the species are culled from the orchard. Likewise when the second *Casuarina* species near sexual maturity, all the females of the species are culled from the orchard. Following culling remaining male *Casuarina* species are allowed to pollinate the remaining female *Casuarina* species. This may be achieved either naturally or using any means known in the
25 field. Provided the orchard is sufficiently isolated, seed produced according to this method should largely be hybrid seed. In one example of the present invention the female *Casuarina* species will be *C. glauca* while the male *Casuarina* species will be *C. cunninghamiana*.

30 In a further refinement of the described method, male and female *Casuarina* species exhibiting desired attributes may also be selected for during culling of the alternate sex plant species. By simply selecting the best individuals within a

family and ultimately by eliminating poorly performing families, superior quality *Casuarina* hybrid species can be selected for.

Within the seed trade industry, hybrid seeds command a pre-eminent position because of their superior vigour, uniformity and performance. Accordingly, the

5 present invention also resides in *Casuarina* hybrid seed produced according to this method. From hybrid seed, *Casuarina* plants may be generated.

Casuarina hybrids produced according to the present invention may be manipulated to enhance their phenotypic or genotypic characteristics. Such techniques for manipulating the hybrids will be known to those of skill in plant

10 breeding and will include cloning, genetic recombination, or further breeding. Thus, the progeny of the initial cross may follow any one of the paths outlined below:

15 1) It may be crossed with either of its parents. Such back crossing would have the effect of enriching the characteristics of that parent in the next generation.

2) It may be crossed with a plant of the same species as one of its parents, either to introduce new characters or to affirm certain characteristics of that species.

20 3) It may be crossed with a plant from a species different from those of either of its parents. Such would have the effect of introducing further new characteristics.

4) It may be crossed with a plant species that is itself the product of one or more crosses.

25 Such breeding programmes are cumulative and will span a number of years, with careful selection taking place at each stage of the programme. By engaging in such programmes new and unique *Casuarina* plant varieties may be generated

30 *Casuarina* plants may be propagated by either direct-seeding into the field or transplanting small bare-root seedlings into the ground. The method for raising small seedlings and subsequently planting bare-root involves the growing of

young seedlings from seed under glasshouse conditions at high densities. These are then moved into the sun and, by regular trimming, sturdy seedlings are produced. These are separated, dipped into a suitable water-absorbent paste or

gel product, and planted directly into the field using appropriate machinery. This method dramatically reduces establishment risk and makes subsequent control of weeds simpler.

Thus, according to a further embodiment of the present invention there is provided

- 5 a method for raising *Casuarina* seedlings comprising the step of: cultivating the seedlings in the presence of a suitable water-absorbent paste or gel product.

In one form of the invention *Casuarina* seed and seedlings are cultivated in the presence of at least a wetted water-absorbent polymers. In general, suitable water-absorbent polymers consist of one or more granular materials that, when

- 10 wetted, convert to a gel or paste form and are capable of absorbing and releasing water repeatedly. Suitable polymers will absorb up to several hundred times their weight in water and can repeatedly absorb and release the water for as long as a decade. When placed near the roots of *Casuarina* plants, such polymers act as "water banks," absorbing excess water until the *Casuarina* plant roots can tap it.
- 15 When properly applied, suitable polymers can reduce the amount of water lost through percolation and evaporation, reduce the leaching of soil nutrients, pesticides, or herbicides, and improve soil aeration. Those effects improve yields and reduce watering cost significantly.

Certain water-absorbent polymers are known to speed the growth of commercially

- 20 valuable plants through improved water management. Polymers that may be used for this purpose include polyacrylate (the absorbent agent used in disposable diapers), polyvinyl alcohol, starch-based copolymers, and cross-linked polyacrylamides. More specific examples of water-absorbent polymer mixes that might be used include, aqueous gels derived from highly absorbent, cross-linked,
- 25 mixed salts of homopolymerised or copolymerised acrylic acid; synthetic agricultural polymers such as polyelectrolytes used in combination with water-soluble polysaccharides; dissolved water-soluble polyisocyanate capped prepolymers; substantially non-ionic polyacrylamides cross-linked with a low amount of methylenebisacrylamide (MBE); or hydro-gel polymers derived from
- 30 polyvinyl alcohol and polyacrylic acid. Blends of such agents and compositions that are commercially available may also be used for this purpose.

If desired, such polymers can also be mixed with soil, nutrient supplement or any other agricultural mixture (eg synthetic growth media) that aids the growth of seedlings.

Where *Casuarina* plants are harvested at repeated intervals, the sustainability of

5 plant growth between the harvest times will depend on the type of soil in which the plants are cultivated and the nutrients and minerals in the soil, as well as a range of other silviculture limiting factors such as whether any disease affects the rootstock through poor cutting techniques, the inability of the plants to sustain regular harvesting, and seasonal impacts on the production of shoots. Those

10 skilled in the horticultural field will recognise that many of these factors can be controlled through the use of fertilisers and minerals, where appropriate, as well as through careful silviculture techniques.

Generally, harvesting will involve cutting individual *Casuarina* stems adjacent to, but above, the roots of the variety. When cut in this manner *Casuarina* plants will

15 re-shoot from the root system left in the ground. Thus, where at least the root system is retained in the ground the plantation may be re-established from the existing roots. Typically, where a 3-year growth and harvest regime is used, this regime is repeated between 1 and 6 times using the same rootstock. Most desirably, the growth and harvest regime is repeated 1 to 4 times and in one

20 example of the invention the growth and harvest regime is repeated 3 times.

The ability of *Casuarina* plants to grow rapidly and at a density of at least about 5,000 plants per hectare provides a significant advantage to other plant species grown in their proximity in that they are able to rapidly establish shelter for those plants, which in turn ensures they are able to grow straight and tall.

25 Thus, according to a second embodiment of the present invention there is provided a method of cultivating timber comprising the steps of:

(a) Selecting at least a plant variety from the plant genus *Casuarina* capable of growth at a density in excess of about 5,000 stems per hectare;

30 (b) cultivating the plant variety at a density in excess of about 5,000 plants per hectare; and

(c) growing at least another, but different species from that employed in step (b) in close proximity to the *Casuarina* plants.

It should be appreciated that given the relative protection provided by the *Casuarina* plantings and the ability of such plants to aid with nitrogen nutrition,

5 any other plant species might be grown in close proximity to the *Casuarina* plantings, depending upon the climatic conditions that pertain to a particular area. Preferably, the species selected and planted will generate relatively "high-value timber" and will take about 10 to 15 years to mature. This provides an opportunity for a financial return, earlier than would otherwise be available if the species
10 selected had to reach maturity before harvesting. Most desirably, the plants should be capable of being harvested at an age of 10 and 15 years as this allows some of the high value trees to be utilised while the best trees grow through to maturity.

A person of ordinary skill in the field of horticulture will recognise that a wide range

15 plants producing high value timber may be employed in the present invention. In a highly preferred form of the invention the "high value timber" producing plants are selected from the varieties: *Grevillea robusta* (silky oak) or *Toona ciliata* (red cedar).

The subject method is not limited to the growth of a single high-value timber
20 species, but may provide for the growth of any number of these species. According to a highly preferred form of the invention, both *Grevillea robusta* and *Toona ciliata* are grown in close proximity to the *Casuarina* plantings. When grown together, both *Grevillea robusta* and *Toona ciliata* may be grown in any ratio. Preferably, however, approximately 80 percent of the crop will be *Grevillea*
25 *robusta*, while about 20 percent will be *Toona ciliata*.

Where more than one high-value timber species is grown, the species may be grown in any planting pattern or combination of patterns suitable for commercial production of the plants. Such planting patterns will be known to those skilled in the growth of such timber. Preferably the high-valued species are widely spaced

30 to encourage diameter growth and avoid the problems of allelopathy (inability of

the species to grow in close proximity). This may be achieved, for example, by spacing the plants in alternating rows.

Irrespective of the planting pattern chosen, the high value timber species should be planted in such a fashion to capture the shelter provided by the *Casuarina*

5 plantings. This might be achieved, for example, by growing the high-value timber species between rows or sets of *Casuarina* plantings.

Pruning, harvesting, and/or thinning of the high value timber species should be carried out at regular intervals to maximise the size and quality of the timber produced. In this respect, removal of branches at various heights during the

10 growth of a tree will allow most trees to develop knot free wood with minimal stem taper.

Harvesting and coppicing the *Casuarina* plants during the growth of the high value timber species will also keep the *Casuarina* plants from competing with the high value timber species. A person skilled in the art will be able to determine a

15 pruning, harvesting, coppicing and thinning timetable, which will produce trees of a desired size and quality.

The use of trees produced as a result of this invention is not restricted in any way. For example, it will be appreciated that the *Casuarina* plants can be harvested for commercial use or used as a relatively renewable source of fuel.

20 BRIEF DESCRIPTION OF THE DRAWINGS

Figure 1 illustrates the positions of *Casuarina* rows relative to plantings of high-value timber species *Grevillea robusta* and *Toona ciliata*. Note the spacings presented in the illustration are provided by way of example only.

They are not intended to depict either the only way of growing these species or the most preferred way of achieving this outcome.

BEST MODE(S) FOR CARRYING OUT THE INVENTION

The features of the present invention are more fully described in the following non-limiting Example. It is to be understood that this detailed described is included solely for the purposes of exemplifying the present invention. It should

not be understood in any way as a restriction on the broad description of the invention as set out above.

Example 1

Positions of plant species

5 Hybrids between *Casuarina cunninghamiana* and *Casuarina glauca* were planted at approximately 40,000 stems/hectare in 2 beds (1 & 2), next to one another, in 1.5 to 1.8 metre (preferably 1.35 metre) wide biomass beds in accordance with Figure 1. The distance between each biomass beds was approximately 1.3 metres and at the head of each row there was a headrow (3). Within each of the 10 biomass beds there are four biomass rows, 400 mm apart with the outside row being 75 mm from the edge of the bed.

Adjacent to each of the biomass beds (1 & 2) high-value timber species (4) *Grevillea robusta* and *Toona ciliata*, were grown. These varieties were planted at a density of 450 stems/hectare and 50 stems/hectare respectively, at 15 approximately 1.3 metres from the beds (1 & 2) in accordance with Figure 1 (see also Table 1).

After approximately 3 years, the *Casuarina* plants (1 & 2) were harvested by cutting the stems near ground level adjacent to the roots. Rootstock left in the ground was allowed to re-shoot, providing a means for re-establishing the 20 *Casuarina* plant rows (1 & 2).

Biomass harvested from the *Casuarina* plants has a wide range of potential uses. One particular use is as a biofuel. *Casuarina* wood is easy to split, has a high calorific value (around 5000 kcal/kg) and burns slowly with little ash. Converting this to MJ/kg (1 kcal/kg = 4.1868 kJ/kg) the calorific value is 20.934 MJ/kg. The 25 energy embodied in wood is largely a function of mass and moisture content. A typical figure used is 20 MJ/kg (IGPO 2001) for dry wood.

After approximately 3 years the *Grevillea robusta* and *Toona ciliata* species were also thinned to reduce their numbers to 225 and 25 stems/hectare, respectively.

Pruning of the *Grevillea robusta* and *Toona ciliata* branches was again carried out after approximately 6.5 years in accordance with Table 1. Briefly, both *Grevillea robusta* and *Toona ciliata* were pruned at various heights to allow the trees to grow tall and straight. Further, harvesting and thinning of the *Grevillea robusta* and *Toona ciliata* was carried out as necessary to reduce competition between the plants.

After a further 3 years (year 6) the *Casuarina* plants were again harvested according to the previous method. Rootstock was again allowed to re-shoot.

In years 7, 8 and 10 the *Grevillea robusta* and *Toona ciliata* were again pruned in accordance with Table 1 to allow the trees to continue grow tall and straight.

Final harvesting of the *Casuarina* plants was undertaken at year 10 in accordance with Table 1. At the same time both the *Grevillea robusta* and *Toona ciliata* plantings were thinned to allow the remaining trees to achieve maximum high and wood quality prior to harvesting at approximately year 15.

15 Table 1 provides a pruning, harvesting, coppicing and thinning regime

Action	<i>Grevillea robusta</i>	<i>Toona ciliata</i>	Hybrid between <i>C. cunninghamiana</i> and <i>C. glauca</i>
Plants per hectare	450	50	40 000
Age 3 Harvest Hybrid & coppice			40 000
Age 3 Prune 250/hectare to 2.2m	225	25	
Age 6.5 Prune 250/hectare to 4.4m	225	25	
Age 6.5 Prune 50/hectare to 6.5m	225	25	
Age 6.5 Thin to waste	225	25	
Age 7 Harvest hybrid and coppice			40 000
Age 7.2 Prune 125/hectare to 8.2m	100	25	
Age 8.1 Prune 125/hectare 10.0m	100	25	
Age 10 Thin 125/hectare	100	25	
Age 10 Harvest hybrid and coppice			40 000
Clearfall harvest 125/hectare	100	25	

Those skilled in the art will appreciate that the invention described herein is susceptible to variations and modifications other than those specifically described. It is to be understood that the invention includes all such variation and modifications. The invention also includes all of the steps, features, compositions 5 and compounds referred to or indicated in the specification, individually or collectively, and any and all combinations or any two or more of the steps or features.

Dated this Twenty Fifth day of January 2002.

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